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IMPACT OF COOKING ON THE FLUORESCENCE OF BOVINE MUSCLES

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I. INTRODUCTION

Red meat has exceptional nutritional properties because it contains all the essential amino acids and heme iron which is easily assimilated by the human body. Most red meat consumed is cooked to increase its digestibility and palatability, but this cooking step can also produce process-induced toxicants (PITs) (1) along with protein oxidation (2) and thermal denaturation (3). In an attempt to characterise the *in situ* changes that take place in red meat following different types of more or less intense cooking, Synchrotron-deep UV fluorescence microspectroscopy was used to acquire multispectral images mapping the changes that take place from the crust to the inside of the piece of meat.

In addition to other analyses enabling a link to be made with the presence of PITs in the samples, this study would make it possible to identify changes in PIT's concentration according to 1) the cooking conditions and 2) the localization in meat sample. These results should pave the way for more relevant mitigation strategies in order to reduce the health impact of the consumption of such toxicants.

II. MATERIALS AND METHODS

Cooking method. Ground beef patties were cooked in a stainless-steel frying pan (17 cm diameter) on a controlled-temperature induction hob (Bosch Electroménager, Saint-Ouen, France) to simulate medium meat (core 70 °C, according to WHO recommendations for ground meats): **OMS**, as well as 2 other more drastic cooking conditions: **11 min** & **15 min**. Temperatures at the core of the meat and at the bottom of the pan were monitored by thermocouples (RS Components, Beauvais, France). Raw and microwaved minced steak have also been formulated to serve as references: **Raw** & **MW**. For each of the 5 conditions, 4 repetitions were done.

DUV Fluorescence Microspectroscopy. Synchrotron DUV microspectroscopy was performed on the DISCO beamline of the SOLEIL synchrotron radiation facility (Saint-Aubin, France). DUV monochromatized light was used at 275 nm excitation wavelength to excite tissue sections. The emission spectra were acquired from 280 to 540 nm. For each cooking condition, emission spectra were acquired in muscle fibres at the surface, at 2 mm from the surface and 4 mm from the surface, as well as from the crust to 4 mm from the crust with at least 20 to 30 points per location. For each condition, a larger mapping was carried out on one of the repetitions.

Spectrum Processing and Statistical Analysis. Autofluorescence spectra were spike- and noise-filtered using an in-house program written in MATLAB, version 7.3 (The MathWorks, Natick, MA). The Orange software (version 3-3.37.0) was used to perform a baseline adjustment to zero, apply unit vector normalization that normalizes sample-wise the spectral data to unit vectors, and analyze the processed spectra by principal component analysis (PCA). Loading plots derived from the first principal components (PC1) and second principal components (PC2) were used to reveal and identify characteristic fluorescence peaks.

III. RESULTS AND DISCUSSION

Each sample was analysed using the same protocol in order to study the variations in fluorescence as a function of cooking conditions and distance from the crust in the muscle fibres.

Looking first at the phenomena occurring at the level of the cooked meat crust and then at greater depths in the different samples, it appeared that microwave-cooked meat behaves in the same way as raw meat, regardless of where it is located in the piece of meat (Figure 1). One possible explanation is related to the fact that micro-wave cooking is known not to produce PITs [4] such as heterocyclic aromatic amines and polycyclic aromatic hydrocarbons, some of which can be monitored using fluorescence. Significant difference in the fluorescence signal at the crust level was found but it disappeared at 2 mm from the surface, where the signals were identical to those found for raw and microwave-cooked meat. These results reflect the existence of physico-chemical phenomena on the surface of a piece of meat during pan-frying.

PCA loadings revealed 3 characteristic emission fluorescence peaks: 1 pic close to 330 assigned to the fluorescence of tryptophan [5], one at 358 nm and another at 403 nm.

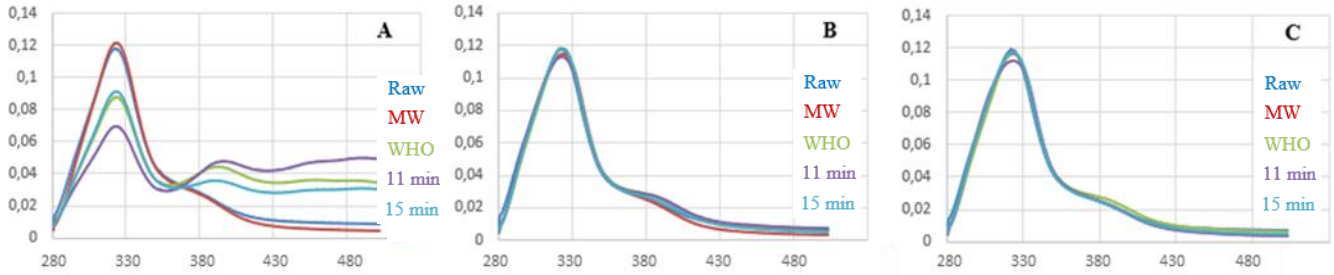


Figure 1. Emission signal corresponding to a 275 nm excitation wavelength found in muscle fibres of meat samples at A) the surface (crust), B) 2mm from the surface and C) 4 mm from the surface. The curves shown correspond to the average values obtained from the 20 to 30 points analysed for each of the 4 repetitions per condition. The values shown are normalized.

In addition to these results, the evolution of the fluorescence signal within a given sample was studied and display in Figure 2. Again, the importance of the location of the measurement in the sample on the fluorescence signal can be highlighted. At the surface, the peak at 403 nm is much greater than inside the piece of meat. When looking at the signals corresponding to the sample cooked in the pan (Figure 2 C), we can see that the deeper we go into the piece of meat, the more the peak at 403 nm decreases and the peak at 330 nm increases until matching the fluorescence signal of raw meat.

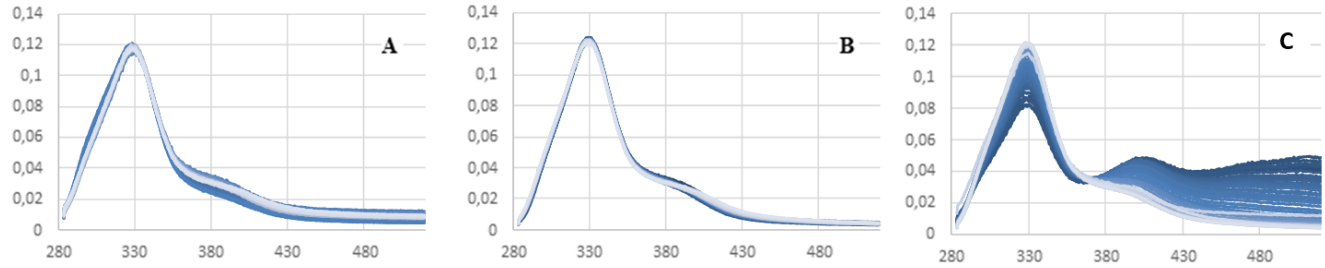


Figure 2: Emission signal found in muscle fibres from the crust (dark blue) to 4mm from the crust (light blue) corresponding to a 275 nm excitation wavelength for A) raw meat, B) meat cooked by MW, C) meat cooked 11 min. The values shown are normalized.

These results show that there is a gradient in the fluorescence signal as a function of distance from the crust of a cooked meat sample. In order to be able to link (or not) these results to the formation of PITs, MALDI imaging is in progress.

IV. CONCLUSION

Based on the fluorescence results of the different cooked meat samples, it appears that fluorescence is directly linked to the thermal process undergone and to the distance from the surface (crust). Most of the physico-chemical phenomena take place at the surface of a piece of meat during pan-frying and the more intense the cooking, the more significant the effect. Further studies are underway to possibly link these variations in fluorescence to the formation of PITs; if this were the case, it would make it possible to develop more efficient mitigation strategies to reduce their impact on human health.

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